

**Department of Pesticide Regulation
Enforcement Branch**

INVESTIGATIVE SAMPLING MANUAL

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INTRODUCTION

This is a manual of standard procedures for the collection of enforcement samples. The purpose of enforcement samples is to provide physical evidence to prove violations of pesticide laws. You should not take samples for purely informational purposes without prior approval from the Department of Pesticide regulation. It is important that you follow the procedures in this manual in order to maintain the integrity of the evidence.

You must determine the goal of the sampling and the appropriate sampling methods which correspond to that goal. Decide what it is you need to prove your case and make a sampling plan to establish that proof.

The nature of the incident will largely determine the way you take samples, if any. You must gather as much background information as possible in order to develop a sampling plan. What and how you sample, how you store and ship samples, and how you document the evidence are all critical to your case. Finally, draw all the elements of the investigation together to prepare your report.

Good sampling procedures and careful investigative techniques will enable you to report your findings with confidence.

1. Sampling Plan

The first step in developing a sampling plan is to review the information you have and to determine whether sampling will prove or disprove the violation. The sampling plan should include the safety precautions, quality assurance requirements, chain of custody, storage and preservation requirements and the number, type, and location of samples based on the requirements of each case. The following outline will help you determine if sampling is necessary and, if so, it will assist you in developing your plan.

Review of available information.

Statements and interview notes. These are an excellent source of information. They will tell you the type of episode; validity of the complaint; urgency of the case; pesticides used; application method; distances between the treatment site and the episode site; weather conditions; date and time of application; application patterns; illness symptoms, and; crop damage patterns.

Records will substantiate the facts gathered during interviews. Records to look for include: pesticide labels; Permits, NOI's and Use reports; recommendations; maps; invoices; work orders, and; field harvest records. A careful examination of the entire episode site along with your review of the

records and interviews will assist you in determining whether violations occurred. Here are some of the things you should look for:

- a) distances from treatment sites.
- b) odors.
- c) crop type, maturity and acreage.
- d) physical surroundings, remember you will be reporting to others who may not be familiar with the area, so make a note of surrounding crops, waterways, landmarks, utility poles, location of workers, vehicles, trees and anything else which could help others to visualize the area.
- e) physical evidence, such as plant damage, tire tracks, spotting on leaves, walls, .
etc., and pesticide containers in storage.

B. Obtain Technical Data from Various Sources

Technical data will help you to determine what type of samples are most appropriate, how to collect them, any time limitations involved, and any special preparation or handling of the samples. Refer to the Pesticide Reference List in appendix D or your supervisor for assistance. Technical data which may affect your sampling plan include:

- a) chemical properties, such as formulation, stability, and metabolites.
- b) physiological and biochemical behavior, such as foliar absorption, translocation, and mode of action.
- c) behavior in or on soils, which includes absorption and leaching, microbial breakdown, oxidation, photo-decomposition, volatilization, and hydrolysis.
- d) toxicity to humans, wildlife, bees; acute and chronic toxicity; and symptoms.
- e) laboratory capabilities including method, sensitivity and standards.

Weather data at the time of the episode, will help you to determine drift patterns and volatilization rates. Weather data include wind speed and direction, moisture, temperature, inversion and humidity.

Sample Selection

Samples can be divided into four types:

- a) Total (expressed as weight of the pesticide/total weight of the sample in ppm).
- b) Dislodgeable (expressed as weight of the pesticide/surface area in μ/cm^2).
- c) Surface (expressed as weight of pesticide/sample μg or weight/area, $\mu\text{g}/\text{cm}^2$).
- d) Volume (expressed as weight of pesticide/volume, mg/mg^3 or $\mu\text{g}/\text{l}$).

Episode Site Diagram

Before you begin sampling, draw a diagram of the episode site. Show the orientation with north at the top of the page. Draw the diagram to scale. Include these things in your diagram: episode site, treatment site, landmarks, crops and acreages, crop stages, room dimensions, witness locations, wind speed and direction.

II. General Sampling Guidelines

Sampling guidelines can be useful even though individual investigations may vary greatly. The sampling plan should include the safety precautions, quality assurance requirements, Chain of Custody, storage and preservation requirements; and number, type, and location of samples based on the requirements of each case. Samples must accurately represent the problem area to justify the effort and expense of analysis. Applying the following guidelines to individual cases will reduce haphazard sampling.

As an investigators should have a sampling plan adapted to the requirements of the individual investigation. You should also have the necessary sampling tools and sample containers. Collect samples **early** in the investigation. The sooner samples are obtained and analyzed, usually, the more meaningful the results. During all phases of the investigation, be careful not to contaminate yourself or cross-contaminate the samples. Always wear clean or disposable gloves, protective clothing, and safety equipment as required by the pesticide label, regulations, and policy when entering the fields or handling samples.

Different types of sample analyses (such as soil to grass) are difficult to compare. Similar materials should be used for comparison samples, such as in cases where treated and nontreated areas are to be compared. Like-foliage types (grass-to-grass) should be sampled when possible.

Also, sampling must be representative of the site and commodity being sampled. Selection of individual sample collection sites must be uniformly distributed (i.e., Nine-Point Grid Pattern) throughout the sample location.

Sample Type

Composite Sample: Combination of two or more sub-samples to produce a homogeneous sample.

Duplicate Sample: One or more samples collected under comparable conditions.

Split Sample: Division of a sample into two equal and identical portions.

Composite sample analyses should only be requested when the area, crop, or material is suspected to be uniformly contaminated, to identify suspected chemicals, or to determine if other samples should be analyzed.

Follow the guidelines under specific sample type to determine sample size. Amounts listed are for single analysis only. Enough material must be submitted to allow for separate analyses if more than one analysis per sample is requested. If composite, duplicate, or split samples are requested, increase the sample size accordingly. Contact the Chemistry Laboratory if in doubt regarding sample size.

8. Identify and officially seal (with custody seals) samples immediately as they are taken.
9. Draw a diagram of the episode site. The diagram should include such things as the sample locations, landmarks, adjacent crops and sites, room dimensions, the location of witnesses, distances between points, etc. Orient diagram to depict North at the top. Be as accurate as possible and include the diagram with the investigation report.
10. **Do not** enclose sample data or Chain of Custody forms in direct contact with sample. Identifying numbers on sample containers should be copied exactly as on the sample data or Chain of Custody Form to identify the sample for the Chemistry Laboratory. Complete the Laboratory's Request for Analysis Form (Appendix A) accurately.
11. Use proper storage and preservation methods. Refer to Section "VI - Sample Storage, Preservation and Shipping" for additional information.
12. Samples should be shipped to the Chemistry Laboratory as soon as possible after they are collected. Basically, samples are best shipped frozen or refrigerated, as appropriate. If this is not practical, the following guidelines may be of some help:
 - a. **Plant material** does well in a ventilated bag.
 - b. **Soils** collected in glass containers should be in heavy plastic bags or double bagged to prevent splitting during shipment.
 - c. **Water** should be shipped in clean glass containers with Teflon or aluminum (foil) lined lids.
 - d. "Blue-Ice" in styrofoam is a good way to ship, especially in the summer.

III. Sampling Equipment

A.

Sampling Equipment List

1. Laboratory Request for Analysis Forms, Chain of Custody Forms
2. Paper and plastic bags (various sizes)
3. Jars (various sizes), foil
4. Labels, tape, stapler, official seals or evidence tape
5. Shovel, handspade, knife, pruning shears, trowel, spatula
6. Leaf punch, isopropyl alcohol or water (according to manufacturer's recommendation), 3-1 oil
7. Sterile pads, cotton balls, isopropanol (pesticide grade), precut templates (made with heavyweight paper)
8. Measuring tape, land measuring wheel

9. Surveyor markers, stakes
10. Camera, film, flash attachment, accessories
11. Ice chests (styrofoam, etc.)
12. "Blue-Ice," wet ice, **dry ice** (**Caution:** should not be handled with bare hands)
13. Permanent markers, grease pen, pencil, pen, note pad, record book
14. Farm map, grower's file, Pest Control Operator's file, county or city map, aerial maps
15. Disposable rubber gloves, rubber gloves
16. Pole with grasping attachment, ladder, net, disposable core tubes, siphon tubes
17. Paper towels, acetone, isopropyl alcohol, water, wash bottle (equipment decontamination)
18. Personal safety equipment and clothing as required by pesticide labels for sample collection (coveralls, respirator, goggles, hard hat, rubber gloves and boots, rainsuit, waders, etc.)

IV. Sampling Procedures

A. Foliage

Pesticide drift, pesticide residue, overtolerance, or other misapplications of pesticides are often documented by sampling and analyzing foliage or whole plants for residues. The sampling techniques described below are applicable for most agricultural crops as well as nonagricultural vegetation (noncrop) where suspected drift or overspray (swath

displacement) is to be documented.

1. Single Foliage Sampling

The following procedures for collecting single foliage samples were written to establish guidelines for collecting other types of foliage samples in this manual. Generally, an investigator would not collect just one foliage sample. Samples are usually collected in patterns (gradient or grid, see Appendices E and F to determine if an area was contaminated). These procedures can be followed whether you sample in a grid or gradient pattern. NOTE: Always wear clean or disposable gloves, protective clothing, and safety equipment as required by the pesticide label, regulations, and policy when entering treated fields or handling samples.

- a. Collect foliage from a specific location (i.e., reference point) with the field. The purpose of collecting the sample from a specific area is to identify a residue delineation between the sample areas, and to maintain sampling uniformity. It is important to identify the location of the sample site within the field, because the sample and episode site diagram may be used as evidence in an administrative or judicial action. The size of the sample area will vary with the type of location. For example:

Field crops and noncrop areas (weeds, fallow fields, etc.) -- the size of the sample site should be approximately a 25-foot square (i.e., 625 square feet).

Orchards and vineyards -- sample from approximately four mature trees/vines (rectangle or square area). The area will vary depending on the size of the episode site, the size of the plants and foliage within the site, and the number of samples intended to be collected.

Smaller plants (seedlings), minimal foliage on a plant or tree (bud leaf stage), or multiple analyses -- a larger area should be used.

Remember, measure the sample area and record it in your investigative notes.

- b. Try to collect foliage of similar type (i.e., grasses or broadleaves only), if possible. It will make it easier to extrapolate the data. If similar type foliage is not available throughout the sampling area, collect different type foliage.
- c. Select foliage from all sides of the plant/tree unless drift is suspected. In this case, collect the foliage from the side of the plants allegedly exposed to the drift. For most situations, collect the foliage from the outer leaves of the plant/tree. It may be necessary to uproot the whole plant if systemic pesticide absorption is suspected. **Do not** select foliage in contact with soil. Remember, new growth not subject to chemical application may affect the results of an analysis.

- d. Collect enough foliage from the specific sample area to equal approximately one pound for each pesticide analysis or screen requested. Place the sample in a clean, unused, double-strength paper bag. The paper bag should be free from print.
- e. Samples should be identified immediately as they are taken. The identification number should be written or taped on the paper bag using an indelible ink pen or permanent marker.
- f. Place the paper bag, with sample, in a plastic bag and seal it with a tie, tape, or official seal. This should prevent moisture from coming in contact with the paper bag and its contents.
- g. Samples should be chilled as soon as possible. An ice chest with regular or "Blue-Ice" can be carried into the field for this purpose.
- h. Laboratory Request for Analysis (Appendix A) or equivalent, and Chain or Custody forms must be filled out in indelible ink and placed in a separate plastic bag containing the sample properly sealed and taped shut. Identify sample material as accurately as possible (i.e., cantaloupes, watermelons, casabas; **not** as "melons"). Also indicate maturity of sample foliage, if applicable. If possible, identify specific pesticides or classes (i.e., organophosphates, phenoxys, etc.) suspected to have caused the problem.
- i. If multiple samples are taken, use new or clean gloves and tools between sampling. Ensure that each sample is identified and that sample material is not contaminated.
- j. Vegetation analyzed for total residue can be stored in a freezer and shipped on dry ice. Ship by fastest method available. If samples are stored frozen, be sure to ship them on dry ice. Otherwise, the plant tissue may decompose during shipment. Refer to Section VI - Sample Storage, Preservation and Shipping for additional information. Maintain Chain of Custody.

2. Grid Pattern Foliage Sampling (Appendix E)

Grid pattern samples are taken to establish uniform or partial contamination of the episode site. The sampling pattern should represent the entire field or site. Each point on the grid represents a sample and should be kept separate from the others. An episode site may be partially contaminated when an applicator does not substantially confine a pesticide to the treatment site (i.e., drift). If pesticide drift is

suspected from an adjacent field, or the source of contamination is unknown, a grid pattern may be used in place of the gradient pattern (refer to 2c.). If misapplication to part of a field is suspected (tank contamination or partial application), but the treated area is unknown, this type of sampling pattern should be used to isolate the area (see Diagram, Appendix F). Wear the required protective clothing and use clean, uncontaminated tools for each sample.

- a. Collect samples in a grid pattern following the procedures indicated for a single foliage sample, as described in Section IV. A.1. Each sample represents one point on the grid for that field or site; therefore, **do not** composite them.
- b. The sampling grid pattern in the episode site should start approximately 100 feet from the edge of the field, depending on the field size. As a rule of thumb, the distance from the edges should represent approximately 10 percent of the width and length of the field or site. For example, a site 1,000 feet wide and 2,000 feet long (approximately 46 acres) has a starting point 100 feet in from the length and 200 feet in from the width.
- c. If you are using the grid pattern to establish drift, collect one additional sample from each of the adjacent fields that are suspected of being the source of contamination. Each sample should be in line with, and at an equal distance apart from, one another in the grid pattern. Record the sample locations in your investigative notes and diagram(s).
- d. If the field or site is suspected of being partially contaminated (drift or three-quarters of the field was treated), start collecting samples from the area that is suspected of containing the least amount of contaminant.
- e. Collect approximately one pound of plant material per sample, per analysis or screen. Be sure to collect enough plant material to accommodate the Chemistry Laboratory if several analyses are requested. Contact the Laboratory if in doubt as to amount of samples to collect.
- f. Each foliage sample from a site should be of similar type and taken from the same location on the plant/tree. On the diagram, identify the location and area from which each sample was taken with distances from landmarks, field borders, and between samples clearly indicated.
- g. Identify each sample as it is taken. Package, preserve, store, and ship samples as described for a single foliage sample in Section IV.A.1. Refer to Section VI

- Sample Storage, Preservation and Shipping, for additional information. Maintain the Chain of Custody.

3. Gradient Foliage Sampling (Appendix F)

Gradient samples are taken to establish a drift pattern. Always sample from an area of suspected lowest concentration in a direction towards the area of highest concentration (treatment site) to prevent contamination of samples. If more than one source of contamination is suspected, collect gradient samples towards each suspected source or use the grid pattern. **Do not** composite samples. Wear the required protective clothing and use clean, uncontaminated gloves and tools between each sample.

- a. Samples should be collected from defined areas in the field or site similar to the methods indicated in the procedures for a single foliage sample as described previously in Section IV.A.1.
- b. Collect a minimum of five foliage samples in a gradient pattern at an equal distance apart. At least one control sample must be from outside of the suspected contaminated area, and one sample must be from the suspected source area of contamination. The gradient pattern should be in a straight line.
- c. Collect approximately one pound of plant material per sample per analysis or screen. Be sure to collect enough plant material to accommodate the Chemistry Laboratory if several analyses are requested.
- d. Foliage should be of similar type and taken from the same location on the plant/tree for each sample. On the diagram, identify the location and area from which each sample was taken with distances from landmarks, field borders, and between samples clearly indicated.
- e. Identify each sample as it is taken. Package, preserve, store, and ship samples as previously described for a single foliage sample in Section IV.A.1. Refer to Section VI - Sample Storage, Preservation, and Shipping, for additional information. Maintain Chain of Custody.

4. Composite Foliage Sampling

Composite samples are taken to determine whether or not an area is contaminated, to determine if other samples should be analyzed and to identify specific chemicals in the sample. The composite sample is made up of several subsamples that are of equal volume or weight and are combined to represent a field or site. Store and ship samples as previously described for a single foliage sample in Section IV.A.1.

Refer to Section VI - Sample Storage, Preservation, and Shipping, for additional information. Maintain Chain of Custody.

If a composite sample is requested, the Chemistry Laboratory will combine foliage from each grid point sample. Remember to collect enough foliage for each grid point sample to allow for the requested analyses.

5. Dislodgeable Foliage Sampling

Dislodgeable foliage samples are collected to determine the potential for exposure of a pesticide to an individual. They are reported in a weight-to- surface area ratio.

Prior to collecting samples, contact your supervisor. Your supervisor can make the arrangements with the Chemistry Laboratory and provide you with the equipment and information necessary to collect the samples. Wear the required protective clothing and safety equipment. **Do not** contaminate yourself.

- a. Dislodgeable samples are taken with a leaf punch device and deposited in an attached clean jar. A sample should consist of forty (40) punches taken with a five-square centimeter punch (large type) or sixty (60) punches taken with a 2.5 square centimeter punch (small type). Leaf punch equipment should be cleaned between each sample using water (or alcohol) and a paper towel.

When punching the leaf, make sure the leaf surface covers the entire cylinder area. A partial leaf punch will result in an inaccurate total surface area.

Measure the punch diameter prior to sampling and include the measurement on the Laboratory Request for Analysis (or equivalent) Form. Record the **exact** number of leaf punches collected per sample.

- b. Select a site where people were working or are likely to come in contact with foliage, but not frequented by human activity. If samples are collected from foliage contacted by people, the pesticide residues may have been dislodged. Sampling sites include a crop in a field, trees in an orchard, shrubs in a residential setting, etc.
- c. Each sample should represent a specific site that can be identified on a diagram. The area represented could be four trees in an orchard, a 25-foot square (i.e., 625 ft.² area) in a row crop, several shrubs, or a tree in a residential setting, etc. The size of the field or orchard, the size of the leaves and plants, and the number of samples to be collected should be considered when selecting a sample site.
- d. The punches should be equally divided between the north/south and east/west sides of the plant to eliminate any effects from differential breakdown. Avoid

taking punches from outside rows as they may not represent the total area being sampled.

- e. Punches should represent all areas of the foliage normally contacted and reachable. This could include the interior as well as the exterior of the plant or tree. **Do not** sample from new growth or leaves contacting the soil unless you suspect they are the source of contamination. If they are the suspected source, be sure to keep soil-contaminated foliage separate from other foliage samples.
- f. Collect enough samples to represent the episode site. For field crops and orchards, the Nine-Point Grid Pattern should be used (see Appendix E). The grid pattern is used to identify contaminated areas (hot spots) in a field caused by drift, tank contamination, etc. A composite sample should be taken if the episode site is **known** to be uniformly contaminated. For a residential setting, two or three samples are usually adequate.
- g. For multiple analyses, sampling should be repeated as described above for **each** analysis or screen requested. Because you cannot sample from the same area, collect companion samples adjacent to each other. There should be a companion sample for every analysis requested. The locations should always be the same size and of the same material. Use a separate jar for each companion sample per analysis and identify with consecutive numbers (JA-7-87-1A; JA-7-87-1B; etc.). The companion samples should represent one sample site. Contact the Chemistry Laboratory to determine if companion samples are necessary. There are situations when one sample (40 punches) is sufficient to analyze for two chemicals.
- h. After collecting a sample, seal the jar with aluminum foil and place the lid on the jar.
- i. Identify each sample as it is taken. Place sample jar in a plastic bag and seal it properly.
- j. Chill immediately; but **do not** freeze. It is recommended to place the jars in an ice chest with wet ice as they are collected. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.
- k. These samples should be given "Priority 1" and marked "Human Health Hazard." Indicate on the Laboratory's Request for Analysis Form that the sample be analyzed for dislodgeable residue. Include the leaf punch size (diameter) and the exact number of leaf punches in the sample.

- l. Ship on wet ice as described in Section VI - Sample Storage, Preservation and Shipping. Maintain Chain of Custody.

6. Commodity Sampling

Commodity samples are collected to determine if pesticide residues are in excess of the tolerance. This information is used to prohibit the harvest of a field, quarantine a packed commodity, etc. It is not used to "clear" a grower's field or for information purposes for a grower.

Field Sampling

- a. Collect single commodity samples in the same manner as you would collect single foliage samples.
- b. Collect commodity samples from the field that are representative of the whole commodity. **Do not** remove wrapper leaves, hulls, shells, pods, etc. Also, **do not** wash or clean the commodity.
- c. If the entire field is suspected of carrying pesticide residues in excess of the tolerance, collect samples in a grid pattern. Collect them in the same manner as you would collect grid pattern foliage samples as described in Section IV.A.2.
- d. As a general rule, collect one pound of commodity per sample, per analysis, or screen. Place the sample in a clean, unused double-strength paper bag.
- e. Identify each sample as it is taken.
- f. Place the paper bag, with sample, in a plastic bag and seal it with a tie (official seal, etc.). This should prevent moisture from coming in contact with the paper bag and its contents.
- g. Samples should be chilled as soon as possible. An ice chest (styrofoam, etc.) with regular or "Blue-Ice" can be used for temporary storage and shipment to the Chemistry Laboratory. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

Packed Sampling

If pesticide contamination of a packed or processed commodity is suspected, it is recommended that the investigator contact his/her supervisor to determine the appropriate State or federal jurisdiction to notify.

General sampling guidelines for packed or processed commodities can be found in the Food and Drug Administration's (FDA) **Inspection Operations Manual**. However, there are some basic points to consider when collecting this kind of sample:

- a. Samples collected at packing sheds should be representative of the produce as shipped in the channels of trade.
- b. Sample size is determined by the number of total units (i.e., cases, crates, boxes, etc.) contained in the lot. The following information can be used as a guideline for determining a "representative" sample size:

<u>Lot Size</u>	<u>Number of Samples</u>
12 or less	5
13 to 18	6
19 to 30	7
31 to 56	8
57 to 190	9
over 190	10

NOTE: Unless otherwise instructed, the minimum subsample size should be two pounds.

- c. **Do not** strip outer leaves before sampling commodity from bulk lots at a packing shed, unless removal of the outer leaves is the practice at the packing shed prior to shipping. Place the sample in a clean, unused double-strength paper bag.
- d. Identify each sample as it is taken. Place the bagged sample in a plastic bag and seal it with a tie (official seal, etc.).
- e. Package, preserve, store, and ship samples as described for a single foliage sample in Section IV.A.1. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

B. Soil

1. Single Soil Sampling (Surface)

Surface soil samples are best for misapplication of herbicides and soil-applied insecticides. A single soil sample is usually one part of the total sampling scheme. They are usually collected in patterns (grid or gradient) to prove an area was contaminated. If the pesticide(s) was incorporated, or otherwise located below the soil surface, then subsurface samples, as described later, should be taken. Always wear clean or disposable gloves and protective clothing, and safety equipment as required by the pesticide label, regulations, and policy when entering treated fields or handling samples.

- a. Collect the surface soil (top one-half inch) from a specific location within the field or site. The sample area should represent approximately a four-foot square (i.e., 16 ft.² area), depending on the size of the episode site, the concentration of the pesticide residue(s), and the number of analyses required. If the episode site is large, the suspected pesticide concentration is relatively low (drift vs. direct application), or if several pesticide analyses are requested, you may want to enlarge the sample area. Measure the sample area and record it in your investigative notes.
- b. Using a clean spatula or other tool, scrape the surface soil down to a depth of one-half inch. Collect enough soil from the premeasured area to fill a clean, one-quart jar with at least one pound of material per analysis or screen. Be sure to seal the glass jars tightly with a Teflon or foil-lined lid.
- c. Samples should be identified immediately as they are taken. The identification number, date, and collector's initials should be written or taped on the sample container using an indelible ink pen. Place jar with sample in a sealed plastic bag. Use official seals if appropriate.
- d. Glass jars should be packed in a manner to prevent breakage.
- e. Mark the location of the sample area on the diagram indicating distances from each sample, landmarks, and field borders.
- f. Samples should be chilled as soon as possible. An ice chest with regular or "Blue-Ice" can be carried into the field for this purpose.
- g. Laboratory Request for Analysis (or equivalent) and Chain of Custody forms must be filled out in indelible ink and placed in a second sealed plastic bag before being placed with the sample.

- h. If multiple samples are taken, use new or clean gloves and tools for each sample to prevent contamination.
- i. Soil samples should be stored, frozen, and shipped frozen on dry ice. Ship by fastest method available. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

2. Single Soil Sampling (Known Depth)

Soil samples at a known depth are collected when the pesticide is suspected of being incorporated, band or rod treated, shanked, trenched, or moved below the soil surface (leaching). If the samples are not collected at the proper depth, the sample analyses will be misleading. This type of sampling will generally be collected in a grid pattern within a field or site. The sampling depth could be 0"-6", 3"-6", 6"-12", etc.

- a. Select a specific sample location and measure an area of approximately one-square foot. The sample area can be enlarged or made smaller depending on the size of the episode site, the area suspected of being treated, the concentration of the pesticide applied, and the number of analyses requested. Record the measured sample area in your investigative notes.
- b. Using a spatula or shovel, remove the soil to the beginning depth you wish to sample. From that point, collect the soil to the desired depth (0"-6", 3"-6", 6"-12", etc.). If a soil probe (e.g., Veihmeyer) is available, remove the soil to the beginning depth you wish to sample (see Diagram entitled Veihmeyer Soil Sampler on page IV-12).

VEIHMEYER SOIL SAMPLER

SAMPLING VARIOUS DEPTHS USING A SOIL SAMPLING TUBE

From that point, take several core samples to the desired depth using the probe. NOTE: It is not recommended to use the probe when a band or side dress treatment was used. Since it is difficult to determine where the band treatment is located, the probe technique could miss the pesticide and give misleading results.

- c. Collect approximately one pound of soil per analysis or screen from the premeasured area and place in a clean glass jar. Seal with a Teflon or foil-lined lid. Glass jars are susceptible to breakage during shipment, so pack them accordingly.
 - d. Samples should be identified immediately as they are taken. The identification number, date, and initials of sampler should be written or taped on the sample container using an indelible ink pen. Place jar with sample in a sealed plastic bag. Use official seals if appropriate.
 - e. Mark the location of the sample area on the diagram indicating distances from each sample, depth of sample, landmarks, and field borders.
 - f. For safety procedures, storage, packing, and shipping of the sample, follow the procedures as described for a single surface soil sample in Section IV.A.1. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.
3. Single Soil Sampling (Known Depth, Furrowed Field)

Chemicals may have been applied in bands or side dressed in furrowed fields. In order to sample from the appropriate area, a shovel is needed to cut across sections perpendicular to the direction of furrow at each sample site (see Diagram entitled Soil Sampling in Furrowed Fields on page IV-14). **Do not** contaminate yourself. Wear clean or disposable gloves, protective clothing, and safety equipment as required by the pesticide label, regulations, and policy when entering fields or handling samples.

SOIL SAMPLING IN FURROWED FIELDS

- a. **Single Rows**: Start at the center of the furrow and sample across the bed to the center of the opposite furrow. Collect soil from an area 3"-6" wide, and 12"-14" deep, as measured from the top of the bed.

Double Rows: If the field is laid out in double row beds, sample from center of furrow to center of bed at 3"-6" width and 12"-14" depth.

- b. Place the soil in a large paper or plastic bag and mix thoroughly. Collect approximately one pound of soil per analysis or screen from the mixed soil and place in a clean, one-quart glass jar sealed with a Teflon or foil-lined lid. Pack samples so the jars are not susceptible to breakage when handling.
 - c. Samples should be identified immediately as they are taken. The identification number should be written or taped on the sample container using an indelible ink pen. Place jar with sample in a sealed plastic bag. Use official seals if appropriate.
 - d. Mark the location of the sample area on the diagram indicating distances from each sample, depth and width of sample, landmarks, and field borders.
 - e. For safety procedures, storage, packing, and shipping of the sample, follow the procedures as described for a single surface soil sample in Section IV.A.1. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.
4. Grid Pattern Soil Sampling

Grid pattern soil sampling is used to establish uniform or partial contamination of the episode site. The sampling pattern should represent the entire field or site. Each point on the grid pattern represents a sample and should be kept separate from the others. There are several pesticides (DDT, DCPA, chlorothalonil, dieldrin) that are long lasting in the environment and adhere to soil particles. Cultural practices (i.e., discing) and wind can move these particles from adjacent fields to a crop, creating a potential residue problem. The grid pattern sampling results can be used to document the transport of the soil particles.

If misapplication to part of a field is suspected, but the treated area is unknown, the grid sampling pattern should be used to define and isolate the area.

- a. Collect soil samples at the appropriate depth in a grid pattern following the procedures indicated for a single soil sample (surface, known depth, furrowed field). Each sample will represent one point on the grid for that field or site. **Do not** composite these samples.
- b. The sampling grid pattern should start approximately 100 feet from the edge of the field, depending on the field size. As a rule of thumb, the closest sample distance from the edges should represent 10 percent of the width and length of the field or site.
- c. If you are using the grid pattern to establish the off-site movement of contaminated soil, collect one additional sample from each of the adjacent fields that are suspected of being the source of contamination. Each sample should be in line with, and at an equal distance apart from, the other samples in the grid pattern. Record the sample locations in your investigative notes.
- d. If the field or site is suspected of being only partially contaminated (i.e., three-quarters of the site was treated), collect samples from the area of lowest concentration to the highest.
- e. Collect approximately one pound of soil per sample, per analysis, or screen.
- f. Mark the location of the sample area on the diagram indicating distances from each sample, landmarks, and field borders.
- g. For safety procedures, storage, packing, and shipping of the samples, follow the procedures as described for a single surface soil sample in Section IV.B.1. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

5. Gradient Soil Sampling

Gradient soil sampling is usually conducted to prove drift or movement off-site from a treated area. Remember that soil samples are of limited use and are not the preferred sampling method in most situations. If drift is suspected to have occurred to a field of plants and the pesticide can be detected in soil, then gradient sampling may be of use. A gradient pattern may also be used to determine if a structural or residential application was made according to label directions.

- a. Samples should be collected from defined areas in the field or site similar to the methods indicated in the procedures for a single surface soil sample as described in Section IV.B.1. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.
- b. Collect a minimum of five surface soil samples in a gradient pattern at an equal distance apart with at least one control sample (collect first) outside the contaminated area, and one sample from the suspected source area of contamination. Collect at least one pound of soil for each analysis or screen requested.
- c. Always sample from an area of suspected lowest concentration in a direction towards the area of highest concentration (treatment site). Collect gradient samples towards each suspected source, if more than one source of contamination is suspected.
- d. Mark the location of the sample area on the diagram indicating distances from each sample, landmarks, and field borders.
- e. For safety procedures, storage, packing, and shipping of the samples, follow the procedures as described in Section IV.B.1. for a single surface soil sample. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

6. Composite Soil Sampling

Composite soil samples are taken to determine if an area is contaminated, to determine if other samples should be analyzed, and to identify specific chemicals in the sample. The composite sample is made up of several subsamples that are combined to represent a field or site.

If composite samples are requested, the Chemistry Laboratory will combine soil from the grid point samples to represent a composite. Collect enough soil for each grid point sample to allow for the requested analyses. Coordinate composite sample requests with your supervisor and the Laboratory Coordinator.

C. Sediment

Single Sediment Sampling

If equipment is not available to collect a sediment sample, or assistance is needed, contact your supervisor. Your supervisor will make the arrangements to have the appropriate agency assist or collect the samples.

Pesticide residues can accumulate in the bottom sediment of lakes and streams. It may be necessary to conduct sediment sampling to document the pesticide source. There are commercially available devices for sediment sampling, but these devices often require extensive cleaning between sampling to prevent cross-contamination. Directly scooping sediment into a glass jar is recommended for shallow sampling situations.

Sediment contents can be flushed or diluted as the jar is lowered or retrieved through water exceeding a few inches in depth. Therefore, a disposable core tube is recommended for unconsolidated sediment, and use of a commercial sediment-collection device is recommended for firm bottom deposits. It is recommended to sample with the flow for shallow-flowing streams.

1. Carefully lower disposable core tube, or other sampling device, through water into sediment (minimize rolling the sediment).
2. Retrieve approximately one pint of sediment.
3. Transfer sediment directly into a clean glass sample jar or a clean pan. Remove rocks, leaves, and other debris from sediment before transferring to the wide-mouth glass jar. Seal with Teflon or foil-lined lid.
4. Label each sample as it is taken, and place the sample jar in a sealed plastic bag. Store on crushed ice. Transport to the Chemistry Laboratory by the most expedient and reliable method. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.
5. Dispose of, or clean, sampling tools. Rinse pan, if used, with isopropyl alcohol and flush with clean water before collecting additional sediment samples.
6. Each sample or subsample location should be identified on the diagram of the incident site.

D. Water

The following guidelines are for collecting samples of surface water, and are designed to detect pesticide residues resulting from some type of misuse (i.e., off-site drift, over spray) of the pesticide to surface water such as lakes, streams, or ponds. If you suspect pesticide contamination of ground water, it is recommended that you contact your supervisor to determine the appropriate local, State, and/or federal agency for follow-up.

Wear shoulder-length gloves and chest-high waders whenever contact is made with potentially contaminated water.

Single Water Sampling

1. Use clean, one-gallon amber glass containers with an aluminum foil or Teflon seal under stopper. Fill bottles to top leaving no air space for pesticides to volatilize.
2. Sample as close as possible to the apparent source of contamination. Avoid areas where water has been isolated from the main body of the stream, lake, or pond.
3. Wade out as far as possible into ponds or streams. Avoid sampling sediment that is disturbed by your movement. Wear shoulder-length gloves and chest-high waders if dermal exposure may be a problem.
4. If the suspected pesticide is water soluble, the sample can be drawn from any depth below 18 inches. If the pesticide is oil-based, or oil is a part of the tank mix and the alleged misapplication was made across the water surface, then the sample can be drawn from the surface layer.
5. Lower the glass bottle to the desired depth with the cap on. Remove the cap under water, and allow bottle to fill. Replace the foil-lined cap and lift the bottle out of the water. For surface samples, dip the bottle into the water surface and allow it to fill completely.
6. Several samples distributed around ponds or lakes are preferable to only one sample. If only one sample is taken, draw several subsamples from different locations around the body of water and combine in a clean, one-gallon container.
7. If the water is too shallow to immerse a jar, use another clean jar to fill the sample jar.

8. Identify each sample as it is taken with an identification number. Place sample jar in a plastic bag and seal it with a tie, tape, or official seal.
9. Refrigerate or place the sample on wet ice immediately. In some cases, other chemicals may be added to the water to aid in preserving the sample. Contact the Chemistry Laboratory performing the analysis for instructions. Additives (i.e., preservatives) must be documented on the Chain of Custody or Laboratory Request for Analysis (or equivalent) forms.
10. Ship sample on wet ice. Pack the sample to avoid breakage and label it adequately. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

E. Air

Generally, volume air sampling is conducted by an environmental or occupational health agency.

Basically, there are two types of air samplers used. High Volume (Hi-Vol) samplers are used to measure low concentrations of pesticides over long periods of time; and Low Volume (Lo-Vol) samplers are used to measure higher concentrations of pesticides over shorter periods of time (see illustration, page IV-21). These are not hard, fast rules, and conditions often dictate which sampler is best.

1. Air Sampling Indoors

Either Hi-Vol or Lo-Vol samplers can be used depending on expected concentrations. NOTE: Air samplers produce a moderate noise. They should be positioned where they will not unnecessarily disturb other persons in the structure, if possible.

- a. Hi-Vol samplers must be vented out of the dwelling to ensure that air will not be recycled through the machine.
- b. Rooms with cigarette smoke or gas appliances must be avoided; any gases or suspended smoke particles in the area will contaminate the sample.

2. Air Sampling Outdoors

Either Hi-Vol or Lo-Vol samplers can be used outdoors.

- a. Sampling equipment must be positioned to avoid exposure to engine exhausts, running motors, cigarette smoke, or any other nontarget air contaminants.
- b. Sampling equipment must also be protected from rain and direct sprays from application machinery. Shelter hoods are used to protect the equipment in such situations.

Since an environmental or occupational health agency usually has the knowledge and experience needed to operate air sampling equipment, it is recommended that they be contacted if volume air sampling is essential.

F. Feed, Milk and Dairy Foods, Eggs

It is recommended that investigators use the sampling protocol of the United States FDA's **Inspection Operations Manual** for proper sample collection of these commodities for compliance (investigational) purposes.

Some state agencies, such as state agriculture departments, may also have established sampling protocols for these types of commodities.

If you suspect pesticide contamination of a feed, milk/dairy food, or egg commodity, it is recommended that you contact your supervisor to determine which appropriate state and/or federal agency to contact for follow-up.

G. Pesticide Formulations.

Sampling pesticide formulations for investigative purposes is necessary to provide evidence of a pesticide misuse, misformulation, product composition, cross-contamination, or other problem. In order for the analytical results of these samples to substantiate a finding that a violation exists, the samples must be representative of the total amount of the material sampled.

Each sampling situation in an investigation must be judged separately as to number, type, and method. However, some general guidelines should be observed:

1. Read and follow all precautionary statements on the label before sampling a formulated pesticide material (i.e., protective clothing and equipment

requirements). If a label is not available, wear the maximum safety equipment (i.e., respirator, coveralls, gloves, boots, and goggles).

2. If possible/practical, collect the entire container with its contents. Smaller samples can always be collected from the larger container at a later date.
3. Since not every ounce of formulated pesticide material can be sampled (e.g., bulk containers) and analyzed, design sampling so all portions of the pesticide are "represented" in the final sample.
4. Bulk containers (dry and liquid materials) should be subsampled. Subsamples of dry materials should be collected from containers in excess of 40 pounds, and for liquid materials from containers in excess of one gallon.
5. Sample tools should be cleaned with acetone and rinsed with clean water or disposed of properly before collecting additional samples.
6. Avoid cross-contaminating samples by keeping these samples separate from all other samples (i.e., residue) at all times.
7. Use care when packaging samples for shipment to the Chemistry Laboratory. Pesticide formulation samples must be packaged to avoid spillage, leakage, or deterioration and the possibility of endangering the safety of persons or the environment. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

For a detailed discussion on specific sampling procedures for a particular sampling situation, investigators should refer to the **EPA Pesticide Inspections Manual, National Enforcement Investigation Center (NEIC) Pesticide Sampling Guide (EPA)** or your state's sampling protocol. It is recommended that you discuss with your supervisor to determine the protocol that is appropriate for the particular situation at hand.

H. Animals, Fish, Honeybees

Animal, Fish, Honeybee Sampling

1. Dead animals, fish, and honeybees must be sampled fresh before decomposition, if possible. Prior to collecting dead animals, contact a governmental veterinarian for proper dissection techniques and appropriate tissue samples. If wildlife is involved, contact a Fish and Game biologist. In

some situations, a governmental veterinarian or Fish and Game biologist will collect the samples. Maintain Chain of Custody.

2. Use disposable gloves when handling animal samples because of the possibility of disease transmission.
3. Small animals and fish should be collected whole and placed in plastic bags. Collect a minimum of four ounces of fresh dead bees or honey and a minimum of one ounce of pollen. Remember to collect enough for each analysis requested. Place sample in a clean, unused jar. Place sample jar/bag in a plastic bag and seal it with a tie or official seal.
4. Chill all animal, fish, and bee samples as they are collected to prevent further degradation. If fish decomposition is evident, upon collection, indicate so on the Laboratory Request for Analysis (or equivalent) Form.
5. Identify each sample as it is taken with an identification number.
6. Freeze and ship all tissue samples as quickly as possible. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

I. Surface (Swab)

Surface or swab sampling is conducted to establish drift, uniform or partial contamination, or the presence of a pesticide on a surface. Surface samples can be taken in patterns, such as in a structure, or in groups to support other sample analyses. Surface sampling should not be used to determine whether or not a hazard exists.

1. Single Surface Sampling

- a. Surface areas can be sampled by using **two sterile gauze pads** moistened with isopropyl alcohol. Areas to be sample may vary in size depending on the estimated concentration of contaminant. Direct application to a surface would not require as large a sample area as drift from greater distances. As a general rule, use a 500 cm square area (20 cm x 25 cm). Smooth "inert" surfaces are the preferred area to sample (i.e., windshield, etc.). However, follow the same methods for sampling uneven surfaces such as rugs, furniture, walls, walkways, counters, etc.
- b. Disposable, heavyweight paper templates can be made from a manila folder and used to delimit the area to be sampled. In situations where a template

cannot be used, carefully measure and outline the area to be sampled. String, pins, tape, or a ruler can be used for outlining purposes. Be sure to decontaminate or use new equipment for each sample. Always record the surface area sampled on the Laboratory Request for Analysis (or equivalent) Form and in your investigative notes.

- c. Before entering the area to be sampled, take a control sample. Moisten **two sterile gauze pads** with the same solvent to be used for the actual sample and place them in a foil-sealed glass jar. **Do not** contaminate the solvent by placing the gauze pad over the mouth of the solvent bottle. While wearing clean or disposable gloves, pour the solvent over the gauze without touching the bottle. Identify the sample as it is taken with an identification number. Complete the Laboratory Request for Analysis Form indicating that the sample is a control. Ship with other samples. Isopropanol (pesticide grade) can be used for both water and oil soluble pesticides.
- d. Select a sample site (i.e., visible residue). Try to avoid areas known to contain waxes. These may interfere with the analysis. Wearing clean or disposable rubber gloves, measure the area to be sampled. The template is the preferred method to measure the area. Always use a new template for each sample. Tape the template to the surface area or measure and outline the area to be sampled.
- e. Use **two sterile gauze pads** moistened with isopropanol or other suitable solvent per sample. Remember not to contaminate the solvent by touching the gauze pads to the solvent bottle. Wipe lightly horizontally across the measured area with the first pad, folding the contaminated portion in, so that a clean surface of the pad is exposed to make another wipe of the area, and continuing until the whole area has been wiped horizontally. Place that pad in a glass jar which has an aluminum foil or Teflon-lined lid (dull side toward the sample). Wipe the area again with the second gauze pad using vertical wipes. Again, fold the contaminated portions of the pad inward. Use the clean outward portions to wipe the whole area. Place the second pad in the same jar as the first pad. Screw the lid on and mark the sample as described below.
- f. Identify the sample immediately. Record the surface area and sample location on the Laboratory Request for Analysis Form, on the incident diagram, and in your investigative notes. Other information that should be recorded on the Laboratory Request for Analysis Form is the solvent used, the suspected ingredients in the pesticide spray solution, and whether the

surface sample represents pesticides in concentrate, tank mix, or drift pattern form.

- g. If multiple analyses are required, the sampling should be repeated on samples from adjacent areas as described above for each analysis or screen requested. There should also be a companion sample for every analysis requested. The locations should always be the same size and of the same surface material. Use a separate jar for each companion sample per analysis and identify with consecutive numbers (i.e., JA-7-87-1A; JA-7-87-1B, etc.). The companion samples should represent one sample site. Seal sample containers with custody seals. Place each sample container in a separate plastic bag and seal it with a tie or official seal.
- h. Store the samples in the freezer and ship on dry ice. Pack the Laboratory Request for Analysis and Chain of Custody forms and samples in the same manner as the soil/foilage samples. Pack the samples to avoid breakage. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

2. Grid Pattern Surface Sampling

Grid pattern samples are taken to establish uniform or partial contamination of the episode site. They can be used to delineate a contaminated site (treatment site) from nontreated sites. For example, it may want to be proven that a broadcast carpet treatment occurred when labeling only allowed a baseboard treatment.

- a. Collect samples in a grid pattern following the procedures indicated for a single surface sample, as described above. Each sample represents one point on the grid for that site. **Do not** composite the samples. Always collect a solvent control sample prior to any other sampling.
- b. If the site is suspected of being partially contaminated, start sample collection in the area of least contamination.
- c. If multiple analyses will be requested, collect companion samples as described previously in Section IV.I.1.g.
- d. Surface samples should be collected from similar surface areas. Identify on the diagram the location and area from which each sample was taken, with distances from landmarks, walls, and between samples clearly indicated.

- e. Identify each sample as it is taken. Officially seal, store, and ship samples as previously described in Section VI - Sample Storage, Preservation and Shipping for additional information. Maintain Chain of Custody.

3. Gradient Surface Sampling

Gradient surface samples are taken to establish a drift pattern in or on a structure. Always sample from an area of lowest concentration in a direction towards the area of highest concentration (treatment site) to prevent contamination of samples.

- a. Samples should be collected from defined areas at the site similar to methods indicated in the procedures for a single surface sample. Always collect a solvent control sample prior to any other sampling.
- b. Collect a minimum of five surface (swab) samples in a gradient pattern at an equal distance apart with at least one sample outside of the contaminated area (if possible), and one sample from the suspected source area of contamination. The gradient pattern should be in a straight line.
- c. If multiple analyses will be requested, collect companion samples as described above for a single surface sample.
- d. Collect samples from similar surface areas. Identify the location on the diagram and the area from which each sample was taken. Include distances from landmarks, walls, and between samples.
- e. Identify each sample as it is taken. Officially seal, store, and ship samples as previously described in Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

J. Clothing

- 1. Be selective when collecting clothing samples. Collect clothing only from people who were allegedly contaminated. Consideration must be given to the type of incident involved. Garments, such as shoes (footwear), could be collected if an applicator was allegedly exposed to a pesticide because of failure to wear protective equipment. Shirts, scarfs, or jackets could be collected if they were exposed to pesticide drift.

2. Clothing samples are usually collected away from the episode site. Should the history of the clothing be unknown, attempt to find out more about the integrity of the sample. Ask the people involved if the clothing has been washed since the incident. If the clothing has been washed, the investigator should not collect the sample unless there are special circumstances which dictate sampling. Consult with your supervisor for guidance if you are unsure.
3. Place each sample in a clean, unused paper bag to prevent cross-contamination, then place the bagged samples in properly sealed plastic bags for shipment. Each sample should be properly identified. Chill samples as they are collected.
4. Inform those people involved that the clothing will not be returned. Have people sign a Release of Clothing Form (Appendix B).
5. When submitting clothing samples for analyses, consideration must be given to the type of episode involved. Clothing samples are collected to determine if pesticide residues are present. They are not used to determine whether the exposure resulted in a health hazard. If the affected area of the clothing is known (i.e., spot caused by spill), the investigator could cut the affected area out and submit it to the Chemistry Laboratory. In some situations, it might be better to submit the entire garment. Indicate on the Laboratory Request for Analysis (or equivalent) Form the area of the clothing the Laboratory should analyze (i.e., shoulder, sleeve, front, etc.).
6. Store the samples in a freezer and ship on dry ice. Refer to Section VI - Sample Storage, Preservation and Shipping, for additional information. Maintain Chain of Custody.

K. Tank Mix

1. Laboratory analysis of tank mix samples will identify the active ingredient and, also possibly, contaminants in the tank mixture. Analysis results can be reported as either percentage by weight or parts per million. The investigator should indicate how the requested results should be reported on the Laboratory Request for Analysis (or equivalent) Form.
2. It is the responsibility of investigators to protect themselves and others during pesticide handling and sampling activities. Tank mixes may be highly toxic. Protective clothing and safety equipment must be used by investigators when sampling. Refer to pesticide labels for precautionary statements. Rubber/Neoprene gloves and goggles/face shield should be

worn when sampling all tank mixes. Wear a respirator, if required by the label, to protect against vapors, dust, powders. If the tank mix ingredients are unknown, assume they are the most hazardous and wear all required safety equipment. Be careful when working around machinery and at busy mixing/loading sites. Be aware of hoses and fittings that may be under pressure, or show signs of leakage. Wash thoroughly with soap and water before eating, drinking, or smoking.

3. Use glass quart bottles or jars with Teflon or propanol-washed, foil-lined lids. **Do not** allow tank mix solutions to contact rubber or plastic as these materials may affect the analytical results. **Do not** use foil-lined lids if the pesticide reacts with metal.
4. Thoroughly agitate liquid in service container or tank.
5. Use a siphon tube and collect a composite sample from three depths: near tank bottom, middle, and near the top of the liquid level. If the solution is adequately mixed to ensure uniformity, a sample can be collected from the drain system. Collect a composite sample from as many points in the drain system as are available. Aerial application rigs can be sampled at spray boom nozzles. Following actual application, loosen boom nozzle and drain the pesticide mix into a glass sample container. Be sure to tighten the nozzles on the boom after you have taken the sample.
6. To reduce the possibility of spillage, **do not** fill the sample jar or bottle above the bottom of the thread line. If the pesticide is highly volatile, fill to the top of the jar to prevent volatilization into the air space.
7. Identify and label each sample as it is taken. Labels or identification should have the following information: sample number, date, contents, and collector's initials. If possible, include a copy of the pesticide label(s) with the sample. Include dilution and mixing directions on the Laboratory Request for Analysis Form. If the label cannot be obtained, also include ingredient statement and other pertinent label information on the Analysis Form.
8. Place each sample in a separate plastic bag and seal it with a tie or official seal. Package each sample separately for shipping. It is best to chill all tank mix samples to prevent degradation. An ice chest with crushed ice can be used to maintain samples below 40°F. Ship by fastest means available, taking into consideration Department of Transportation (DOT) regulations. Refer to Section VI - Sample Storage, Preservation and

Shipping, for additional information. Maintain Chain of Custody.

9. Decontaminate all tools and change or wash gloves to prevent cross-contamination of samples. **Do not** store or ship tank mix samples with or near foliage or soil samples.
10. After collecting the samples, wash thoroughly with soap and water.

V. Completing the Laboratory Request for Analysis Form (Appendix A)

The Laboratory Request for Analysis (or equivalent) Form should be completed as accurately as possible to ensure the rapid analysis of sample material. Failure to complete the form may result in a delay at the Laboratory.

Any sample has the possibility of becoming evidence in an administrative or judicial action. It is important to properly identify samples in the field and record all the information required on the form. This includes the Chain of Custody information.

Always use a separate data sheet for each sample, companion sample, or subsample submitted.

NOTE: The following is a step-by-step procedure for completing a typical analysis request document. Although the information is specific for one state agriculture department's analysis request document, it is representative of the kind of information that is often needed by the Chemistry Laboratory. You should consult with the Chemistry Laboratory conducting the analyses of your samples regarding the specific information, as well as the accepted format they require with each sample.

1. **Laboratory Number and Date Received:** The Chemistry Laboratory will enter their own identification number and the date upon receipt of the sample.
2. **Agency Name and Address:** Enter the name and address of the agency submitting the sample. The form will be returned to this address with the analysis results. The phone number and name of a contact person should be entered here or below.
3. **Check if Custody Record is Required:** Check this box if the Chain of Custody must be maintained. Be sure to complete the Record of Custody section on the reverse of the form.
4. **Sample Consists of:** Be specific when completing the description of the sample contents (i.e., leaves, twigs, fruit, dead bees, etc.). If the sample is a commodity, give the specific name. If multiple subsamples are sent together, identify each one as accurately as possible. If the sample is not adequately described, the Laboratory may perform the wrong analysis or the analysis may be delayed until confirmation is received from the submitting agency. For dislodgeable samples, the number and area of the punch size should be recorded. As much information as possible should be given for tank mix samples. Include identification and approximate percentages of any fertilizers, sticker/spreaders, buffers, and active ingredients in the mix. Also give brand names or active ingredients, if known. Samples of spilled tank mixes or concentrates taken by the swab method will result in higher analysis results than normal. Report this sample type on the form so that the Laboratory can use different analysis methods. Always list the type of solvent used when taking a swab sample. The Laboratory can supply solvents upon request.

5. **Sample Identification Marks:** Submit a separate form for each sample or subsample. The identification number on the sample must correspond to the identification number on the Laboratory Request for Analysis Form. If not indicated by separate identification numbers and forms, the Laboratory may assume that the samples are a composite when, in fact, they are individual subsamples. The Laboratory will assign its own identification numbers to each sample when it is received. The identification number entered on the Laboratory Request for Analysis Form should be logical and consecutive, especially in the case of subsamples. The investigator's initials should usually be part of the identification number. Ink may run or bleed if used directly on commodities such as watermelons. Either carve the number into the rind of large fruit or include an identification number sealed in an envelope attached to the sample.
6. **Location/Source of Sample:** A brief, but accurate, description of where the sample was taken. Distances from landmarks and field borders can be used. Coordinated numbers may be used to locate subsample locations.
7. **County:** County in which sample was taken.
8. **Detailed Description of Problem:** In the space provided, give a detailed description of the problem. If the sample was taken for compliance purposes only, insert "NA."
9. **Signature, Title, and Date:** Person requesting analysis should sign and date the form.
10. **Sample Priority:** Review the criteria for priority on the back of the Laboratory Request for Analysis Form and check the appropriate box. Routine samples will be analyzed on a first-come, first-served basis, and in order or priority.
11. **Basis for Sample:** Check the appropriate box. This information helps place the sample in the proper section of the Laboratory.
12. **State Specific Analysis Requested:** Be as specific as possible. Request an individual chemical when suspected. Screening analyses are available for some chemical groups and should be indicated when desired. Results for tank mixes or concentrates are given in percentages, unless otherwise requested.
13. **Laboratory Findings:** Results for residues will be given in Parts Per Million (PPM) unless otherwise requested. Dislodgeable results will be reported in weight-to-surface area. Surface sample results will be reported in weight-to-sample weight ratio (ug/sample).
14. **Requested Disposition of Remaining Sample:** Give the Laboratory instructions on what to do with portions of the sample that are not used or destroyed in the analyses. Samples will

- be discarded unless other instructions (i.e., "save sample," "save extracts") to the contrary are given. Request that nonperishable samples be returned to the agency requesting the analysis, if the length of storage is prolonged. Agencies may also consider sending only a portion of the evidence (sample) collected to the Laboratory and maintain custody of the balance at the agency level where suitable storage conditions can be maintained.
15. Results Phoned: Results are sometimes phoned to the agency requesting analysis. It is important to list a contact person and phone number either with the agency name or in the Detailed Description of Problem (number eight) section of the form. The Laboratory Coordinator will usually phone the results to the agency. Laboratory Request for Analysis forms can also be faxed if necessary; however, the original will also always be mailed to the agency.
 16. Chemist and Date: The Laboratory Supervisor or chemist performing the analysis will sign and date the form.
 17. Record of Custody for Requester Sample Number: Enter the identification number again and sign and date the Record of Custody section. Every time the sample changes hands, the signature of the person receiving the sample, the name of the person from whom received, and the date should be entered. If the Record of Custody is incomplete, the Laboratory cannot legally verify the resulting analysis because of the unknown history of the sample.

VI. Sample Storage, Preservation, and Shipping

The proper collection, storage, and shipping of samples are all critical elements of the sampling process and can affect the analysis results. Steps must be taken early in the sampling process to avoid anything that could compromise the integrity of the sample, such as loss, contamination, or tampering.

Ideally, samples should be analyzed as soon as possible after they are collected. However, in many situations, this may not be possible and consideration must then be given to assuring the integrity of the sample by utilizing proper storage, preservation, and shipping methods.

A. Storage, Preservation

If samples must be stored temporarily, it is best to refrigerate or even freeze them to prevent deterioration of the sample and degradation of the chemical. Contact your supervisor if in doubt about the specific requirements for storage or a particular kind of sample.

The table on page VI-2 summarizes the general requirements for preserving various sample types during storage (i.e., field and laboratory) and shipping to the Chemistry Laboratory. Refer to the specific sample type in the "Sampling Procedures" section of this manual for additional information.

B. Shipping

Packaging and shipping samples must be done properly to ensure that they remain **intact** when they arrive at the Chemistry Laboratory. Any loss of the sample can have a negative impact on the admissibility of the sample as evidence. In addition, the safety of persons can be endangered because of loss through spillage, leakage, or deterioration of samples.

Prior to shipment of samples, contact the Chemistry laboratory coordinator or your supervisor to ensure that the Laboratory has the analytical capabilities for the pesticides requested and that the samples are shipped properly, picked up on arrival, and convey special analysis instructions, if any.

SAMPLE PRESERVATION

Sample Type	Container	Field	Storage	Ship
FOLIAGE				
Single	PB	C	F	DI
Grid	PB	C	F	DI
Gradient	PB	C	F	DI
Composite	PB	C	F	DI
Dislodgeable	GJ	C	R	WI
Commodity (field, packed)	PB	C	R	WI
SOIL				
Single (surface)	GJ	C	F	DI
Single (known depth)	GJ	C	F	DI
Single (furrowed field)	GJ	C	F	DI
Grid	GJ	C	F	DI
Gradient	GJ	C	F	DI
Composite	GJ	C	F	DI
SEDIMENT	GJ	C	F	DI
WATER	GJ	WI	R	WI
AIR	GJ	DI	F	DI
FEED, MILK/DAIRY FOODS, EGGS	Refer to appropriate protocol	Refer to appropriate protocol	Refer to appropriate protocol	Refer to appropriate protocol
PESTICIDE FORMULATIONS	GJ	COOL	COOL	COOL
ANIMALS,FISH,HONEYBEES	GJ	C	F	DI
SURFACE				
Swab	GJ	C	F	DI
Grid	GJ	C	F	DI
Gradient	GJ	C	F	DI
CLOTHING	PB	C	F	DI
TANK MIX	GJ	C	R	WI

NOTE: All sample containers are to be sealed in a plastic bag in the field and placed in a second plastic bag with sample data sheets and custody forms.

Codes: **C** Chill (Wet Ice or "Blue-Ice") **R** Refrigerate
DI Dry Ice **PB** Paper bag/sealed in plastic bag
F Freeze **WI** Wet Ice
GJ Glass jar/sealed in Plastic Bag

The following general guidelines are to be observed when packaging samples for shipping:

1. Place properly bagged and labeled samples in shipping container and immobilize samples with suitable packing material such as crumpled newspaper or styrofoam. Seal the shipping container.
2. Pack liquid samples in sufficient absorbent material to absorb and retain any leakage that might occur.
3. Samples to be analyzed for pesticide residues (foliage, soil, water, animal tissue, etc.) require that a temperature be maintained during shipping that will prevent deterioration. Refer to the Sample Preservation Table on page VI-2.

Frozen samples should be placed in dry ice, wrapped in newspaper and placed in an insulated container such as a styrofoam cooler. The insulated container is then placed inside a suitable shipping carton with adequate ventilation provided.

Samples requiring only that they be kept cool should be packed in an insulated container using wet ice or "Blue-Ice."

Keep all liquid sample containers separated and carefully padded to guard against breakage.

4. Clearly mark shipping container with handling instructions, such as "Handle with Care," "Glass," "This Side Up," or other appropriate wording.
5. Comply with all applicable packaging and shipping requirements of DOT.
6. Ship or deliver the samples to the appropriate location as soon as possible and maintain Chain of Custody.

VII. Examples

A. Direct Application Pesticide Misuse

Based on the evidence gathered, it may be determined that there was an alleged misuse of a pesticide on a commodity or at a site. The types of possible misuses are: use inconsistent with the labeling; crop not specified on the label; increased rate; increased concentration; a frequency more than specified, or early reentry, etc. This could result, or have resulted, in an overtolerance on the commodity, a contaminated site (i.e., golf course, park, etc.), or potential human exposure.

The objectives of collecting samples are to prove a violation occurred (FIFRA Section 12.136j) and/or whether there are illegal residues on the produce in the field so that a Stop Harvest Order can be issued. Another objective is to determine if a field is safe to enter by field workers.

1. Sampling Plan

Outline sampling goals and plan. Consider the type of pesticide and its chemical properties, method of application, time, etc., before sampling is initiated. Refer to the "Sampling Plan" section in this manual.

2. Sample Collection

This manual outlines the sampling procedures for different types of sampling situations that are likely to be encountered. Prior to collecting samples, refer to the Sampling Procedures Section of this manual to determine the method, location, size, and handling precautions.

- a. Collect at least nine foliage (whole, leaf/blade) samples in a grid pattern, starting approximately 100 feet from the edge of the field, depending on field size (see Grid Pattern Examples, Appendix E).
- b. Collect at least nine soil samples in a grid pattern. Soil samples should be collected if the pesticide was soil injected, side dressed, broadcast preplant/preemergent, etc. The sampling procedures depend on the pesticide's chemical properties, method of application, placement of the pesticide (broadcast, band, side dress), etc. Consider whether the pesticide could be found on foliage, the commodity, or primarily in the soil. Remember, soil samples are taken to substantiate that a violation occurred.
- c. If the crop is close to harvest, collect at least nine commodity samples in a grid pattern. Commodity samples should be collected in the same manner as harvested, not the way it is packed or offered for sale to the consumer.

- d. Collect at least nine dislodgeable leaf samples in a grid pattern, if the field caused, or may cause, an illness to field workers. If workers were only in one section of the field, sampling should be concentrated in that area. Dislodgeable samples are collected to determine the potential for exposure of a pesticide to an individual and are reported in a weight-to-surface area ratio. Collect samples only if workers may come into, or have been in, contact with vegetation with dislodgeable residues. Dislodgeable pesticide residues are those that originated from a **foliar** application and dislodged through contact with the plant. Pesticides that are soil injected or side dressed normally would not be dislodged from the plant.

B. Pesticide Drift

During a drift investigation, it may be necessary for the investigator to collect samples to determine if drift occurred. It is also important to document what actually occurred through interviews and statements. This information will help in developing a Sampling Plan.

The objectives of collecting samples are to prove that the applicator did not substantially confine the pesticide to the application site and/or whether there are illegal residues on the produce in the field so that harvest can be stopped. Also, another objective is to determine if a field is safe to enter by field workers.

1. Sampling Plan

Review records in the office (i.e., permits, grower maps, Notices of Intent to Apply Pesticides, use reports, application records, etc.) to assist you in understanding the situation. The drift could have occurred from an application several weeks prior to the initial complaint. Also, review notes from interviews and statements. There are times when the source of contamination cannot be identified, but can be narrowed down to two or three sources.

Once the alleged source(s) of contamination is determined, an outline of the Sampling Plan can be prepared. Consider the pesticide and its chemical properties; method of application; timing; wind speed and direction, if known; direction of application; etc. Refer to the "Sampling Plan (Section I) in this manual for further information.

2. Sample Collection

This manual outlines the sampling procedures for the different types of samples. Prior to collecting samples, refer to the "Sampling Procedures" section in this manual to determine the method, location, size, and handling procedures for sampling.

- a. Collect a minimum of five foliage (whole leaf/blade) samples in a gradient pattern. Start sampling from an area of lowest suspected concentration in a direction towards the area of highest concentration (treatment site). Collect like samples at an equal distance from each other. The number of samples in the gradient pattern will depend on the distance between the treatment site(s) and the complaint (i.e., illness, odor, crop damage). Consider the number of pesticides to be analyzed. Collect enough foliage for multiple analyses. If the drift is suspected to have originated from a field adjacent to the episode site, a grid pattern may be used in place of the gradient pattern (see specific procedures under "Grid Pattern Foliage" Sampling section).
- b. Soil samples should be collected only if foliage is not available. Pesticides generally break down more readily in soil because of microbial action, heat, leaching, and photodecomposition. It can be difficult for the Chemistry Laboratory to extract some pesticides because of their binding action with soil particles.

Collect a minimum of five surface soil samples in a gradient pattern. Start sampling from the suspected lowest area of concentration and proceed in a direction towards an area of highest potential concentration (treatment site). Collect the soil samples from the surface (top one-half inch) of a measured area, at an equal distance apart. The number of samples will depend on the distance between the treatment site(s) and the complaint (i.e., illness, odor, or crop damage).

- c. Surface (swab) samples can be collected to determine if a pesticide is present on a nonporous surface. Collect surface samples from vehicles, windows, irrigation pipe, signs, or any other nonporous surface. Always collect one control sample prior to any other sampling collection. Collect at least three samples from the area most likely to be contaminated. If a vehicle passed through the alleged drift, attempt to collect samples from the contaminated area (i.e., windshield, etc.), including the inside of the vehicle if the windows were open. Measure the surface area and be sure to include this information on the Laboratory Request for Analysis (or equivalent) Form.
- d. Clothing samples may be collected to determine if a person was contaminated by pesticide drift. Because clothing samples are usually collected off the episode site, the history of clothing may be unknown and the samples could lose their integrity. Obtain as much information as possible for each clothing sample collected.

Clothing samples should be selected carefully. Collect clothing from people who were allegedly contaminated. Obtain the owner's signature on the Clothing Release Form and inform them that their clothing will not be returned.

- e. Dislodgeable leaf samples are collected to determine the potential for exposure of a pesticide to an individual in the treated field. Dislodgeable residues are reported in a weight-to-surface area ratio.

Collect several samples in the area where people were allegedly exposed. If possible, use a grid pattern within the field or site. If a grid pattern is not possible, select locations where the workers or public were present, but not frequented by them. Because the drift could have occurred several days prior to the exposure, a field worker crew may have worked in the contaminated area.

- f. Collect at least three commodity (i.e., whole fruit, vegetable, nuts, etc.) samples from the field(s) suspected of being contaminated. Select an area that you suspect would contain the **highest level** of pesticide residues. The commodity samples should be collected whole, not the form in which it is packed or offered for sale to the consumer.

C. Structural Pesticide Misuse: Direct Application/Movement Off-Site

Based on the evidence gathered, a determination was made that there was an alleged misuse of a pesticide in or around a structure. The types of possible misuses are: use inconsistent with labeling, site not on the label, increased rate, increased concentration, etc. These misuses could result in a complaint or illness.

The objective of collecting samples is to prove a violation occurred (FIFRA Section 12, 136j).

1. Sampling Plan

Outline sampling goals and plan. Consider the type of pesticide and its chemical properties, method of application, labeling prohibitions, etc., before you start sampling. Refer to the "Sampling Plan" section in this manual.

2. Sample Collection

This manual outlines the sampling procedures for the different types of samples collected. Prior to collecting samples, refer to the "Sampling Procedures" section to determine the method, location, size, and handling procedures for sampling.

a. Inside a Structure

Most labels for structural use pesticides list very specific sites that can be treated. Whether it is crack and crevice, broadcast, baseboard, or spot treatment, determine where the applicator was required to apply the pesticide(s). This information can only be obtained from the label or the applicator. Compare this site and the alleged treated site with the site on the label and determine if there are any conflicts. In order to prove misuse or drift, the sampling pattern should be designed to show a delineation between the treatment site(s) and the suspected off-target site(s).

- (1) Collect one control surface (swab) sample prior to any contact with the contaminated area.
- (2) Collect several (three or more) surface samples in areas suspected of being contaminated from direct application or off-target movement of the pesticide(s). If possible, a grid or gradient pattern should be used to delineate pesticide contaminated from noncontaminated areas. Start from an area of suspected least concentration. The sample area could include carpet, furniture, walkways, vents, walls, floors, etc. However, nonporous surfaces such as metal, glass, or tile will yield the highest recoveries of suspected pesticides.
- (3) Collect one surface sample from the area supposedly, or known to be, treated. If it is impossible to reach the area to collect the sample for suspected misuse from a crack and crevice treatment, collect your last sample from the area closest to the treatment site.
- (4) Draw a diagram of the episode site. Include your sampling pattern, the treatment site, the location of furniture, fixed walls, and doors, sampled surfaces and areas, distances between samples, and reference points, etc.

b. Soil Applied Treatment (Dursban, Diazinon)

Pesticides registered for use to control termites and other pests also have very specific sites that can be treated. Broadcast, perimeter of the foundation, a five-foot band of soil around the foundation of the structure, subslab area (rod treatment), and an eight-inch deep trench around each pillar and the foundation perimeter are some of the sites listed on the labels. Determine where the applicator was required to apply the pesticide(s) and compare this site and the **alleged** treated site with the site on the label and

determine if there are any conflicts. In order to prove misuse or drift, the sampling pattern should show a delineation between the labeled treatment site(s) and the off-target site.

- (1) Collect several (three or more) soil samples in areas suspected of being contaminated from direct application or off-target movement of the pesticide(s).
- (2) Collect surface soil samples to prove off-target movement (drift) or misuse of a broadcast treatment.
- (3) Collect soil samples at a specific depth if the pesticide was trenched, rod-treated, or moved below the surface in some way.
- (4) Collect one or more soil samples at the appropriate depth(s) from the area supposedly or known to be treated.
- (5) If applicable, collect several (three or more) foliage or surface samples in areas suspected of being contaminated from direct application or off-target movement of the pesticide(s).
- (6) Draw a diagram of the episode site and include the location of samples, distances between samples and landmarks, the area suspected of being treated, etc.

APPENDICES

- A. Laboratory's Request for Analysis Form
- B. Release of Clothing Form
- C. Equipment Sources
- D. Pesticide Reference List
- E. Grid Pattern Examples
- F. Episode Sampling Diagrams
 - 1. Crop Damage/Residue Episode
 - 2. Human Episode
 - 3. Property Loss Episode
 - 4. Agricultural Pesticide Misuse
 - 5. Structural Pesticide Misuse